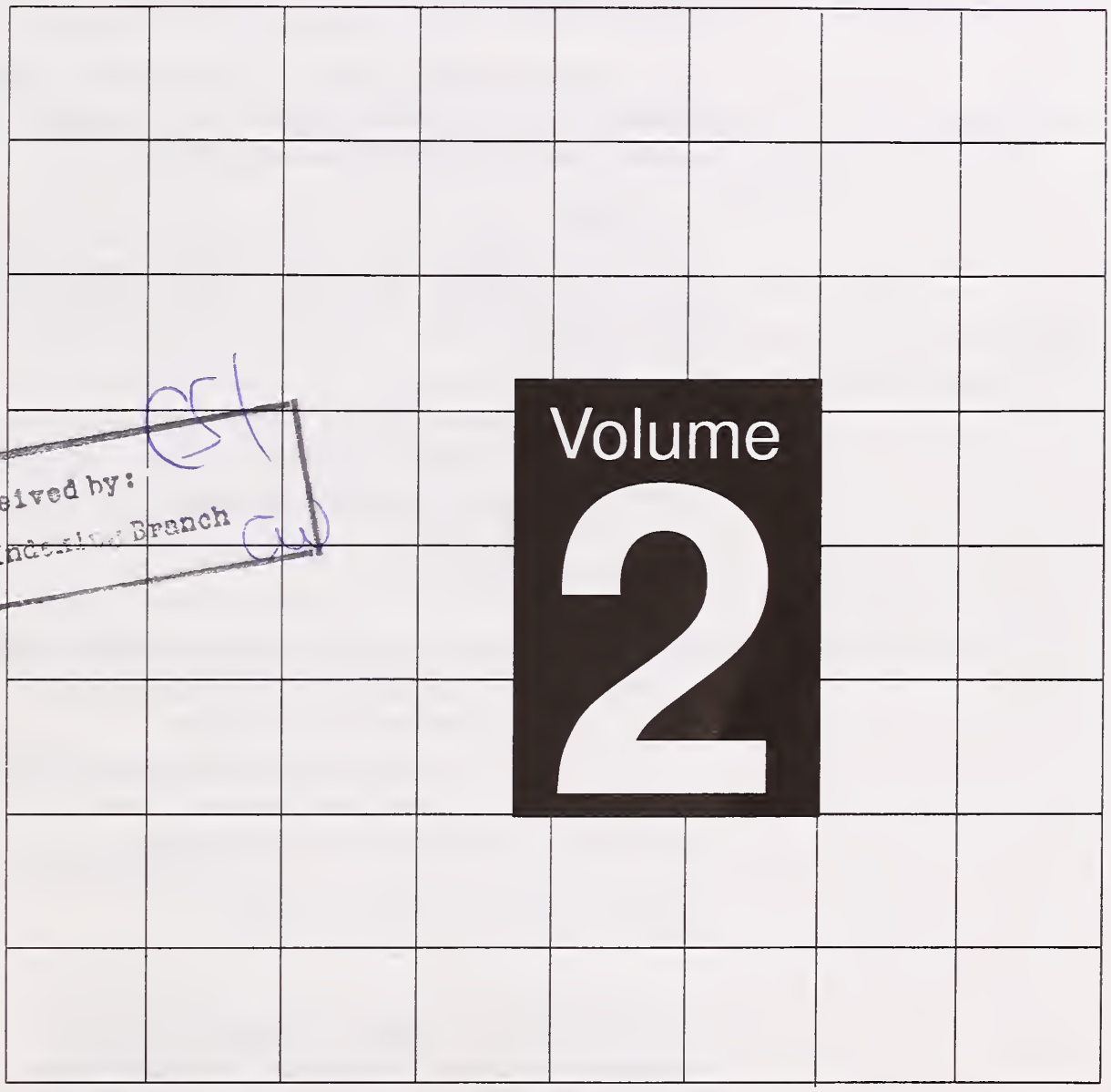
United States
Department of
Agriculture

Animal and
Plant Health
Inspection
Service

Program Aid 1577

Keeping America Free From Foreign Animal Diseases

African Swine Fever



Guidelines for Using This Package

This binder contains an integrated suite of educational materials about African swine fever. The package can be used in a formal training setting, where a presenter will show the video tape and narrate the slide show using this black-and-white brochure as the script. Or the materials can be used in a self-study program with the reader progressing at his or her own pace.

Within this brochure, readers will notice that certain paragraphs are preceded by a number. These numbers correlate to the slide set. For example, the African swine fever slides are all marked "SF" at the top of each plastic slide frame and numbered sequentially from 1 to 54.

If you remove the slides from their protective clear-plastic sleeve (for example, to put them into a carousel for group viewing), please be sure to reposition them in the correct numeric order for the benefit of future users.

This shrink-wrapped suite includes a general and a scientific video tape on African swine fever, a separate slide set on that disease, and the brochure you are reading now. If your package is incomplete, please contact the following office for replacement materials:

U.S. Department of Agriculture
Animal and Plant Health Inspection Service
Veterinary Services, Emergency Programs
4700 River Road, Unit 41
Riverdale, MD 20737–1231

Instructional packages on other diseases are also available and may be requested by writing to the above address. Titles include
Program Aid 1576 African Horse Sickness
Program Aid 1578 Contagious Bovine Pleuropneumonia
Program Aid 1579 Lumpy Skin Disease, Sheep Pox, Goat Pox
Program Aid 1580 Malignant Catarrhal Fever
Program Aid 1581 Rinderpest, Peste des Petits Ruminants
Program Aid 1582 Vesicular Diseases

Issued April 1997

1 African Swine Fever

Definition

2 African swine fever (ASF) is a tick-borne, contagious, febrile, systemic viral disease of swine caused by an unclassified (iridoviruslike) virus.

Etiology

3 ASF is caused by a large (about 200 nm), lipoprotein-enveloped, icosahedral double-stranded DNA virus.

4 Electron micrograph of ASF virus (ASFV) showing low and high magnification of a virion attached to an erythrocyte. In the high magnification, the distinctive shape of the virion and its capsule can be seen.

For many years, ASFV was classified as a member of the family Iridoviridae. But recently, the agent was found to have many characteristics of the poxviruses. Thus, researchers have suggested establishment of a new family for ASFV.

ASFV is quite stable. It will survive:

- Over a wide range of pH. (In serum-free medium, ASFV is inactivated at pH 3.9 or lower and at pH 11.5 or higher. In the presence of 25-percent serum, ASFV persisted for 7 days at pH 13.4.)
- 15 weeks in putrefied blood
- 3 hours at 50 °C
- 70 days in blood on wooden boards
- 11 days in feces held at room temperature
- 18 months in pig blood held at 4 °C
- 150 days in boned meat held at 39 °F
- 300 days in salted dried hams

Effective Disinfectants

5 Effective disinfectants include iodophore disinfectants, chlorine disinfectants, and those containing o–phenylphenol.

Over the years, lower virulence isolates of ASFV have emerged, particularly in the Iberian peninsula. Today, therefore, the virulence of ASFV infections varies from high (causing essentially 100-percent mortality in 7 to 10 days after exposure) to moderate (causing acute illness with a high percentage of pigs surviving) to so mild that only seroconversion takes place.

History

6 In 1921, Montgomery described outbreaks of ASF that occurred between 1909 and 1915 in Kenya in which 1,352 of 1,366 infected pigs (98.9 percent) died. He initially thought the disease was a variant of hog cholera. In 1921, Montgomery also reported that the disease was caused by a virus and gave the results of transmission studies and discussed virus survival in the environment, host range, and the role of wild pigs in maintaining the virus in nature.

Until 1957, ASF was confined to Africa. In that year, it was diagnosed in Portugal. The disease was probably introduced via meat products from Portugal's African colonies. This introduction of the disease is believed to have been eradicated.

In 1960, ASF was again introduced into Portugal and spread throughout that country and to Spain. By 1995, ASF had been eradicated from domestic pigs in both of these countries. Between 1960 and the 1980's, however, ASF spread to additional countries outside Africa, probably as a result of feeding pigs uncooked garbage containing pork that originated in Spain or Portugal.

This calendar documents those ASF outbreaks occurring outside Africa:

1964, 1967, 1974: France
1967: Italy—ASF is still present in Sardinia.
1971: Cuba
1978: Brazil, Dominican Republic
1979: Haiti

1980: Cuba
1983: Italy(mainland)
1985: Belgium
1986: Netherlands

Except in Sardinia, ASF has been eradicated from all these countries.

ASF is still endemic in several African countries.

Host Range

7

At one time, domestic and wild pigs (in Africa, warthogs, bush pigs, and giant forest hogs; and in Europe, wild pigs) were thought to be the only hosts of ASFV.

In 1963, however, Spanish workers isolated ASFV from the soft tick *Ornithodoros erradicus* collected from ASF-infected farms. Subsequently, researchers showed that ASFV replicated in the tick and that there was transstadial, transovarial, and sexual transmission in *Ornithodoros* ticks. *O. moubata* collected from warthog burrows in Africa were shown to be infected with ASFV. ASFV in the wild pigs in Africa is now believed to cycle between the soft ticks living in warthog burrows and newborn warthogs.

Ornithodoros ticks collected from Haiti, the Dominican Republic, and southern California have been shown to be capable vectors of ASFV. But, in contrast to the African ticks, many of the ticks from California died after being infected with ASFV. Many researchers now believe that ASFV is really a tick virus and that the pig is an accidental host.

Because ASFV-infected ticks can infect pigs, ASFV is the only DNA virus that can qualify as an arbovirus.

8

Photograph of a partially engorged *Ornithodoros* tick.

The warthog and bush pig develop viremia but have only very mild or subclinical disease. The disease causes high rates of mortality in domestic pigs and wild European pigs.

Geographic Distribution

9 ASF is present in several African countries and on the island of Sardinia.

Transmission

10 Even though the soft tick has been shown to be a vector (and in Africa probably the reservoir) of ASFV, the primary method by which ASF has spread from country to country has been through the feeding of uncooked garbage containing ASFV-infected pork scraps to pigs. Once a pig becomes infected, ASFV spreads by direct contact, carrier pigs, contaminated people, equipment, vehicles, and feed. The role of carrier pigs has been difficult to prove experimentally, but circumstantial evidence from the field incriminates carrier pigs. An outbreak of ASF in a contained swine operation in Africa was traced to workers feeding the entrails of guinea fowl to pigs. It was shown that the guinea fowl fed on ticks; hence, there was ASFV in the guinea-fowl intestines fed to the pigs.

The amount of ASFV needed to infect a pig depends on the route of exposure. In experiments, a pig can be infected by intramuscular or intravenous inoculation with 0.13 hemadsorbing dose $(HAD)_{50}$. Intranasal–oral inoculation requires 18,200 HAD_{50}.

Epidemiology

11 In areas where ASF is endemic and where there are soft ticks, ticks can be the source of infection. However, in such areas in Africa, pigs can be very successfully raised in confinement with proper isolation and sanitary procedures. The production method with the highest risk of ASF in Africa is the "village pig" system, where pigs roam about freely or, if confined, are not kept in isolation.

In other areas, ASF has to be introduced via infected live pigs or by feeding uncooked garbage containing ASFV-infected pork. Once introduced into a herd, the disease will spread by direct and indirect contact with secretions and excretions from infected pigs. Aerosol transmission is not important in the spread of ASF. Because ASFV does not replicate in epithelial cells, the amount of virus shed by an ASF-infected pig is much less than the amount of virus shed by a hog-cholera-infected pig. The blood of a recently infected

pig contains a very high ASFV titer—$10^{5.3}$ to $10^{9.3}$ HAD_{50} per mL. Therefore, if pigs fight and lose blood, if an infected pig develops bloody diarrhea, or if an infected pig is necropsied, there is massive environmental contamination.

Over the years, lower virulence isolates of ASFV emerged in the Iberian peninsula. Many pigs infected by a lower virulence ASFV isolate survive infection. Two to 3 weeks after infection, these pigs will have a normal temperature and appear normal but still be viremic. If these pigs are introduced into a clean herd, they could be a source of infection; if the pigs were slaughtered for meat, their tissues could be a source of infected meat scraps in garbage.

ASFV will persist in recovered pigs for a long time. In an experiment, pigs became infected with ASFV when inoculated with supernatant from spleen, lung, and lymph nodes collected from pigs about 1 year after an acute ASF infection. These apparently recovered pigs had no gross lesions.

Piglets born of ASF-convalescent dams are free of ASFV and ASF antibody at birth but seroconvert after ingesting colostrum. When piglets from noninfected (control) and from ASF-convalescent dams were challenge-inoculated at 7 weeks of age, the control piglets developed an average viremia of $10^{7.6}$ HAD_{50}/mL of blood and died, while the piglets from convalescent gilts developed an average viremia of $10^{4.9}$ HAD_{50}/mL of blood and survived. When farmers in Cameroon repopulated their herds using ASF-convalescent animals, the herds experienced recurring periods of high mortality due to ASF.

Incubation Period

12 After intranasal–oral exposure to ASFV, pigs develop fever and leukopenia in 48 to 72 hours.

Pathogenesis

13 Other than a bite by an infected tick, the most common portal of entry in natural ASFV infection is the mouth. The virus then most likely enters the tonsils, replicates in local lymphoid tissue, and spreads throughout the body via the blood.

More specifically, ASFV replicates in selected cells of the mononuclear-phagocytic system, and virions attach to erythrocytes. In pigs that survive an ASFV infection, viremia gradually decreases to a nondetectable level by in-vitro isolation procedures over a period of 5 to 8 weeks. Epithelial cells are not infected.

Pigs infected by a highly virulent isolate of ASFV have extensive necrosis in some lymphoid tissues. When they die, 7 to 10 days post infection (DPI), they have no detectable antibody and their tissues are positive by direct immunofluorescence. In contrast, pigs infected by ASFV isolates of less virulence do not have extensive necrosis in lymphoid tissue. These animals develop immunoglobulin M (IgM) about 4 DPI and immunoglobulin G (IgG) about 6 to 7 DPI. Their tissues become negative by direct immunofluorescence about 6 to 7 DPI (near normal immune response).

Research directed toward explaining this difference in antibody response found that ASFV infects Langerhans cells and interdigitating dendritic cells, both of which are antigen-presenting cells.

14 Photomicrograph of normal swine Langerhans cells.

ASFV isolates of high virulence kill the swine Langerhans cells; isolates of low virulence do not. The difference in antibody response is therefore most likely due to the fact that infection of antigen-presenting cells by a highly virulent isolate kills the cell before antigen is processed and presented, while infection by an isolate of low virulence allows normal cell function to continue.

By in-situ hybridization, researchers also found that ASF DNA production continued until death in pigs infected by a highly virulent isolate, while in pigs infected by an isolate of lower virulence, production of ASF DNA started to slow at 4 DPI and essentially stopped by 6 to 7 DPI.

One significant difference between ASF and most viral diseases is that in ASF, IgG does not completely neutralize ASFV in the conventional virus neutralization test and does not clear the viremia.

Pigs that recover from an ASF infection are resistant to reinfection by the same isolate. By knowing the virulence of isolates, researchers, by exposing pigs to increasingly virulent live ASFV isolates, are able to build up the pigs' resistance to the highly virulent ASF isolates.

Clinical Signs

Highly and Moderately Virulent ASFV Infection

The clinical signs of ASF are influenced by the virulence of the ASFV isolate and the physiological state—age, pregnancy status—of the pig. The most common types of ASFV that have caused disease in the field are related to the highly virulent Lisbon 60 isolate (an African virus introduced into Lisbon, Portugal, in 1960) and to a related, moderately virulent isolate (e.g., Dominican Republic isolate).

Following inoculation of feeder pigs with either a highly virulent or moderately virulent isolate, the clinical course for both isolates is similar for the first 4 to 6 DPI. About 2 DPI, the pigs will develop a fever of 105 to 107 °F (40.5 to 41.7 °C), moderate anorexia, and leukopenia. White pigs will have a reddened skin. When disturbed, the pigs will get up and move around. But if left alone, after a short time they will lie down. After 4 to 6 DPI, there will start to be a difference in clinical signs between pigs inoculated with the different isolates.

15 **Infection by a Highly Virulent ASFV Isolate**—Infected pigs become progressively more sick—eat and move less—and most die between 7 and 10 DPI. It is not unusual to see a pig walking and a short time later find it dead.

16 Graph of the temperature and leukocyte counts during the course an infection by a highly virulent (Lisbon 60) ASFV isolate. The course of the disease is short; initially there is a high fever and leukopenia and then death.

17 **Infection by a Moderately Virulent ASFV Isolate**—Infected pigs usually have a high fever for 10 to 12 DPI. Some mortality usually occurs at this time. After 12 to 14 DPI, temperatures and leukocyte counts start to return to normal levels. It is not unusual to have one or more pigs die as early as 7 to 8 DPI, but when these pigs are necropsied, the cause of death is frequently hemorrhage into the stomach. The underlying causes of death were that ASFV infection caused a thrombocytopenia and thus a prolonged bleeding time, and there was a preexisting gastric ulcer. Very young pigs may have high mortality rates and have lesions similar to those caused by infection by highly virulent virus.

18 Graph of the temperature, leukocyte counts, virus isolation from the blood, and IgG antibody in a pig infected by a moderately virulent (Brazil) isolate of ASFV. There is a high fever and leukopenia. The temperature and leukocyte count return to within the normal range about 12 DPI. Virus is isolated from the blood from 2 to 24 DPI. Anti-ASF IgG is detected starting 10 DPI.

Pigs affected with either a highly or moderately virulent ASFV isolate, in addition to developing reddened skin, may develop dark red to purple discoloration of the skin on the ears, tail, extremities of the legs, or skin on the hams. This is a nonspecific sign also seen in other diseases. Some groups of pigs will develop diarrhea, probably due to disturbed gut physiology and flora rather than to a direct effect of the virus because ASFV does not replicate in epithelium. In contrast to pigs with hog cholera (classical swine fever), ASFV-infected pigs do not develop conjunctivitis or encephalitis. Also, despite their high fever, ASFV-infected pigs stay in good condition, whereas pigs with hog cholera quickly lose a lot of weight.

19 A reddened tip of an ear.

20 Reddened skin over the hams.

21 Pigs about 18 days after infection by the Brazilian isolate of ASFV. Except for wanting to lie down, the pigs appeared normal.

Pregnant animals infected with a high-, moderate-, or low-virulence ASF isolate abort.

Clinical Signs of a Low-Virulence ASFV Infection

22 Nonpregnant animals infected by certain low-virulence isolates of ASFV may seroconvert but not show any other sign. Pregnant animals will abort.

Other low-virulence ASFV isolates will cause pigs to have a low fever for 2 to 3 weeks and then to develop reddened areas of skin from 1 to many square centimeters in size. These reddened areas then become raised and necrotic. The pigs may also have painless enlargements of joints, particularly the carpal and tarsal joints. This form of the disease is chronic ASF. Many of these pigs will have recurring episodes of a more acute disease and eventually die during an acute episode.

Gross Lesions

In Highly Virulent ASFV Infection

Pigs that die peracutely from an infection with a highly virulent ASFV may have poorly developed lesions. Animals that die 7 or more DPI have more-classic lesions. Three lesions most consistently found and highly suggestive of ASF infection are

- A greatly enlarged, dark red to black, friable spleen,
- Very enlarged, hemorrhagic gastrohepatic lymph nodes, and
- Very enlarged, hemorrhagic renal lymph nodes.

Other lesions described for ASF are more variable:

- Red to purple areas of skin on ears, feet, and tail,
- Petechial hemorrhages on serosal surfaces,
- Petechial to ecchymotic hemorrhages in the renal cortex,
- Perirenal edema,
- Edema of the gall bladder,
- Swollen liver, and
- Edema of the lung.

In pigs infected orally, the submandibular lymph node may be enlarged and hemorrhagic. Other peripheral lymph nodes may have only edema.

In Moderately Virulent ASFV Infection

The gross lesions 8 to 12 DPI in pigs infected with a moderately virulent ASFV resemble lesions in pigs infected by a highly virulent ASFV. The main difference in the lesions between these two is that, in infections by a moderately virulent ASFV, the spleen, though enlarged, has a more normal color and is not friable.

Photographs of Gross Lesions in Pigs Infected by Highly or Moderately Virulent ASFV

23 Enlarged, dark spleen. In an infection by a highly virulent ASFV, the spleen is greatly enlarged, dark, and friable as a result of extensive necrosis in the spleen.

24 Comparison of a spleen from an ASFV-infected pig with a spleen from a hog-cholera-infected pig. The spleen from the hog-cholera-infected pig is normal in size and color.

25 Greatly enlarged dark (hemorrhagic) gastrohepatic lymph nodes.

26 Enlarged dark (hemorrhagic) renal lymph nodes. Note that there is also perirenal edema.

27 Severe petechial to ecchymotic hemorrhage in the kidney.

28 Edema of the gall bladder and congestion of blood vessels.

In Low-Virulence ASFV Infection

The most common lesions in chronic ASF are necrotic skin lesions, consolidated lobules in the lung, generalized lymphadenopathy, swollen joints, and pericarditis.

Photographs of Gross Lesions in Chronic ASF

29 Reddened, raised areas of skin.

30 Closeup of a more advanced skin lesion. The reddened, raised area has become necrotic.

31 Skin lesions have progressed to the point that the necrotic areas are detaching.

32 Reddened, raised area of skin behind the ear.

33 Necrotic areas of skin on the abdomen.

34 Consolidated lobe of lung.

35 Cut surface of consolidated lung.

36 Enlarged mediastinal lymph node with generalized lymphadenopathy.

Aborted fetuses may be anasarcous, and there may be a mottled liver and petechial hemorrhages in the placenta, skin, and myocardium.

Microscopic Lesions

In infections by a highly virulent ASFV, there is extensive necrosis of cells in the mononuclear-phagocytic system. This is best seen in the spleen, Kupffer cells in the liver, and the paracortical areas of lymph nodes. In moderately virulent ASFV infections, the same cells are affected but do not become necrotic.

Photomicrographs

37 Section of spleen from a pig infected by a highly virulent ASFV isolate. The red pulp is packed with erythrocytes; the pale pink, homogeneous areas are necrotic macrophage-sheathed arteries. Note the persistence of lymph nodules.

38 Higher magnification of the previous slide, showing necrotic macrophage-sheathed arteries.

39 Direct immunofluorescent staining of lung from a pig infected with a highly virulent ASFV isolate.

40 Direct immunofluorescent staining of kidney from a pig thus infected.

41 Direct immunofluorescent staining of spleen from a pig infected with a highly virulent ASFV isolate. Note that most of the ASF antigen is in the marginal zone of the periarterial lymphoid sheath.

42 Higher magnification of the previous slide. Central artery and fluorescence in the marginal zone.

43 Section of reddened skin. There is thrombosis of the subcutaneous vessels.

Morbidity and Mortality

44 Morbidity in a previously unexposed herd will usually be 100 percent in pigs that have contact with each other. Mortality varies with the virulence of the isolate. Highly virulent isolates will cause about 100-percent mortality. Infection by less virulent isolates can cause mortality that varies from a low percentage to 60 to 70 percent. Factors that can increase mortality in infections by the less virulent isolates are other diseases in the pigs, a young age, and pregnancy.

Diagnosis

Field Diagnosis

45 The highly virulent form of ASF will be easiest to suspect because essentially 100 percent of affected pigs will die. ASF caused by the less virulent isolates will be more difficult to diagnose. ASF should always be suspected when there are febrile pigs and necropsy findings include

- A greatly enlarged, dark red to black, friable spleen,
- Very enlarged, hemorrhagic gastrohepatic lymph nodes, and
- Very enlarged, hemorrhagic renal lymph nodes.

ASF has been frequently misdiagnosed as hog cholera. In contrast to pigs with hog cholera, ASFV-infected pigs do not develop conjunctivitis or encephalitis. Also, despite their high fever, the ASFV-infected pigs stay in good condition, whereas pigs with hog cholera are severely depressed and quickly lose a lot of weight. Pigs with hog cholera usually have foul-smelling diarrhea. When hog cholera was endemic in the United States, practitioners said they could diagnose it by that odor.

Specimens for Laboratory

46 ASFV is present in the blood from about 2 DPI until death in pigs infected by highly virulent isolates of ASFV. In animals infected by less virulent isolates, ASFV can usually be isolated from the blood for 25 or more DPI.

The following specimens for laboratory diagnosis should be submitted refrigerated or frozen:

- Heparinized blood,
- Clotted blood or serum,
- Submandibular lymph node,
- Inguinal lymph node,
- Tonsil,
- Spleen,
- Gastrohepatic lymph node,
- Lung,
- Liver, and
- Kidney.

These samples should be submitted in formalin:

- Pieces of the preceding tissues,
- Brain, and
- Any other gross lesion.

Aborted fetuses are usually free of virus; therefore, it is necessary to submit a blood sample from the dam.

Laboratory Diagnosis

47 The initial diagnosis of ASF in an area where it is not known to occur requires isolation and identification of the virus. After the initial diagnosis, it can be confirmed by demonstrating ASF antigen in tissue or ASF antibody.

48 Virus isolation. There are two primary ways to isolate ASFV: primary buffy-coat cultures and pig inoculation.

When the macrophages in a buffy-coat culture become infected, erythrocytes in the culture adhere (hemadsorption) to the infected macrophages to form what is called a rosette.

49 Photomicrograph of a stained rosette.

50 Photomicrograph of an unstained rosette.

Pig inoculation is more sensitive than buffy-coat cultures. In experimental work, buffy-coat cultures have been negative after three blind passages of a specimen, but the same material inoculated into pigs caused ASF. Because of confusion with hog cholera, the classic scheme for pig inoculation to diagnose ASF has been to inoculate naive and hog-cholera-vaccinated pigs.

Antigen Detection

51 ASF antigen can be detected by the direct immunofluorescence test on frozen tissue sections. Direct immunofluorescence was one of the most common tests to diagnose hog cholera. When using the test to diagnose ASF, one must be aware of the differences between ASF and hog cholera.

In ASF:

- Tissues from pigs infected by a highly virulent isolate will usually be immunofluorescent-positive throughout the course of the disease.
- Tissues from pigs infected by a less virulent isolate may be immunofluorescent-positive between 2 and 7 DPI.
- The tonsil is not a tissue of choice for ASF diagnosis.

Serology

52 Antibody can be detected by any of several tests:

- Indirect immunofluorescent test,
- Enzyme-linked immunosorbent assay (ELISA),
- Immunoelectro-osmophoresis (IEOP) test,
- Indirect immunoperoxidase plaque test, or
- Western blot.

When performing laboratory tests to confirm a suspected case of ASF, it is important to examine specimens for both ASF antigen and antibody because in infections by less virulent isolates of ASFV, the specimens may have been collected after the antigen has disappeared from the tissues, but there will be ASF antibody. In a study in Spain, if the specimens were tested only for antigen, 60 percent to 70 percent of the cases of ASF were confirmed; however, if the specimens were tested for both antigen and antibody, more than 99 percent of the cases were confirmed.

Vaccination

53 There is no vaccine.

Control or Eradication

54 Introduction of ASF into free areas can be prevented by

- Cooking all garbage fed to pigs (commercial and backyard pigs, as well as pets such as potbellied pigs), and
- Importing only ASF-free pigs.

Control and eradication of ASF in developed countries can be accomplished by

- Slaughter and disposal of all acutely infected pigs,
- Widespread testing and elimination of all seropostive animals, and
- Good herd isolation and sanitary practices.

Today, ASF is not nearly as great a threat to the United States as it was several years ago, for the major pork-exporting countries have eradicated the disease from domestic pigs.